Positive effect of erythrocyte-delivered dexamethasone in ataxia-telangiectasia

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ABSTRACT

Objective: Ataxia-telangiectasia (AT) is a rare, devastating neurodegenerative disease presenting with early-onset ataxia, oculocutaneous telangiectasia, immunodeficiency, radiosensitivity, and proneness to cancer. In a previous phase 2 study, we showed that 6 monthly infusions of autologous erythrocytes loaded with dexamethasone (EryDex; EryDel, Urbino, Italy) were effective in improving neurologic impairment in young patients with AT. The present article reports the results of the extension of this study for an additional 24-month period.

Methods: After the end of the first trial, 4 patients continued to be treated with monthly EryDex infusions for an additional 24 months, and their clinical outcome was compared with that of 7 age-matched patients who stopped the treatment after the first 6 infusions. The protocol included serial assessment of ataxia (by International Cooperative Ataxia Rating Scale) and adaptive behavior (by Vineland Adaptive Behavior Scales) and clinical and laboratory tests revealing treatment- and steroid-dependent adverse effects, if present.

Results: Patients in the extended study experienced a continuous neurologic improvement with respect to their pretreatment status, whereas controls showed a progressive neurologic deterioration (according to the natural history of the disease) after the discontinuation of the treatment. The delivery system we adopted proved to be safe and well-tolerated, and none of the side effects usually associated with the chronic administration of corticosteroids were observed during the entire trial.

Conclusions: These promising preliminary results call for a large-scale controlled study on protracted treatment of patients with AT with dexamethasone-loaded erythrocytes. *Neurol Neuroimmunol Neuroinflamm* 2015;2:e98; doi: 10.1212/NXI.0000000000000098

GLOSSARY

AT = ataxia-telangiectasia; **ATM** = ataxia telangiectasia mutated; **DSP** = dexamethasone phosphate; **ICARS** = International Cooperative Ataxia Rating Scale; **VABS** = Vineland Adaptive Behavior Scales.

Ataxia-telangiectasia (AT) is a rare genetic disease[1,2] caused by mutations in the ataxia telangiectasia mutated (*ATM*) gene, which results in a multisystemic disorder presenting with early-onset ataxia, oculocutaneous teleangiectasias, progressive supranuclear ophthalmoplegia, immunodeficiency, recurrent sinopulmonary infections, radiosensitivity, and proneness to cancer. In the classic form, patients are wheelchair-dependent by the age of 10 years,[1] and their life expectancy is approximately 25 years.[1,2] No effective disease-modifying therapy is presently available.

Observational studies have suggested that betamethasone may be effective in improving neurologic functions in patients with AT.[3,4] A controlled short-term trial confirmed the efficacy of

Table Demographic data, diagnostic characteristics, and response to treatment

Patient ID	Sex	Age, y/mo	ATM genotype	ATM protein	ICARS Δ score 0–7[a]	ICARS Δ score 0–30[a]	ICARS Δ score 7–30[a]	VABS Δ aa 0–7[a]	VABS Δ aa 0–30[a]	VABS Δ aa 7–30[a]	DSP mean dose 1–6[b]	DSP mean dose 7–30[b]
Continuers												
02-01	M	8/8	6679C>T/8484del A	10%	−4	−15	−11	0/8	1/5	0/9	4.1	8.6
02-02	M	8/7	5979del5/7408T>G	20%	3	−2	−5	0/4	0/9	0/5	2.4	9.0
02-05	M	14/2	IVS12+1 G>T/3576 G>A	ND	−2	−17	−15	1/2	6/1	4/9	17.5	10.1
02-08	M	10/2	6679C>T/6679C>T	Absent	−7	−9	−2	1	3/5	2/5	4.2	10.2
Discontinuers												
01-01	F	11/7	ND	Absent	−12	6	18	2/9	1	−1/9	9.6	ND
01-02	F	8/2	ND	Absent	−16	5	23	2/10	1/10	−1	8.5	ND
01-04	F	10/8	7517del4/?	Absent	−11	7	18	2/3	0/8	−1/7	18.6	ND
01-05	M	10/1	717delCCTC/717delCCTC	Absent	−7	−3	−4	3	1/5	−1/7	3.5	ND
01-07	M	10/2	97delC/2113del T	Absent	−1	16	17	4/4	−1/6	−5/10	1.9	ND
01-10	F	19/2	4396C>T/9139C>T	Absent	−12	14	26	1/3	0/1	−1/2	17.4	ND
01-11	F	13/2	3802delG/?	Absent	−17	2	19	1/6	1/2	−0/4	14.5	ND

Abbreviations: ATM = ataxia telangiectasia mutated; DSP = dexamethasone phosphate; ICARS = International Cooperative Ataxia Rating Scale; ND = not done; VABS = Vineland Adaptive Behavior Scales.
[a] Variations of total ICARS score/VABS average age (aa; expressed in y/mo) in subsequent assessments. 0: baseline assessment; 7: assessment performed after the 6th infusion; 30: assessment performed after the 30th infusion in continuers (4 patients) and 24 months after the interruption of the treatment in discontinuers (7 patients). Ataxia improvement is expressed by a reduction of ICARS score.
[b] The mean dose of DSP of all the infusion bags for each patient (mg/bag).

oral betamethasone (0.1 mg/kg),[5] but corticosteroid side effects were quickly observed. To overcome these side effects and explore the effects of a more protracted treatment on neurologic outcome, we recently performed a single-arm, open-label, 6-month extended phase 2 trial in which the enrolled patients received a monthly treatment of 50 mL of autologous erythrocytes loaded with 2 vials of 250 mg of dexamethasone phosphate (DSP) by EryDex (EryDel, Urbino, Italy) procedure.[6] Similarly to betamethasone, dexamethasone has a high anti-inflammatory potency and lacks mineralcorticoid activity. DSP is converted to dexamethasone by erythrocytes' phosphatases and released into the bloodstream for about 20–30 days.[7]

After 6 infusions, patients experienced a clinical improvement of about 5 points on the International Cooperative Ataxia Rating Scale (ICARS)[8] and a significant improvement in adaptive behavior, assessed by Vineland Adaptive Behavior Scales (VABS),[9] without the occurrence of steroid-dependent adverse events. The improvement was more relevant in patients presenting with less severe neurologic impairment and was affected by the efficiency of the DSP loading procedure into erythrocytes.[6] We report the results of the extension of this preliminary study for an additional 24-month period.

METHODS At the end of the first 6-month trial,[6] 4 male patients (mean age 10.6 years; SD 2.8 years; range 8.7–14.2 years) requested and were authorized by the Ethic Committee to continue EryDex treatment. So far they have been treated monthly for an additional 24-month period. The ICARS and VABS scores of the patients in the extended study before the beginning of the treatment and after 6 and 24 EryDex infusions were compared with those of the 7 patients with AT (2 male, 5 female; mean age 11.8 years; SD 3.5 years; range 8.2–19.2 years) who stopped the treatment after the sixth infusion, as originally planned by the study design.[6] In accordance with a drug compassionate use clause, the selection of the continuers was based on the parents' decision (i.e., no external inclusion criterion could be applied), and the controls were selected among discontinuers by trying to best match the clinical characteristics of the continuers (table). During the extension study, the loading procedure was further optimized by obtaining an encapsulated DSP of 9.4 ± 0.8 mg/bag (vs 6.7 ± 6.9 in the previous study[6]) with the same dose of the medicament. The assessment also included clinical and laboratory tests aimed at detecting treatment- and steroid-dependent adverse effects.

Standard protocol approvals, registrations, and patient consents. The extension was authorized by the Ethic Committee. Current Controlled Trial registration 2010-022315-19SpA.

Figure Neurologic impairment variation and duration of the treatment in patients with ataxia-telangiectasia
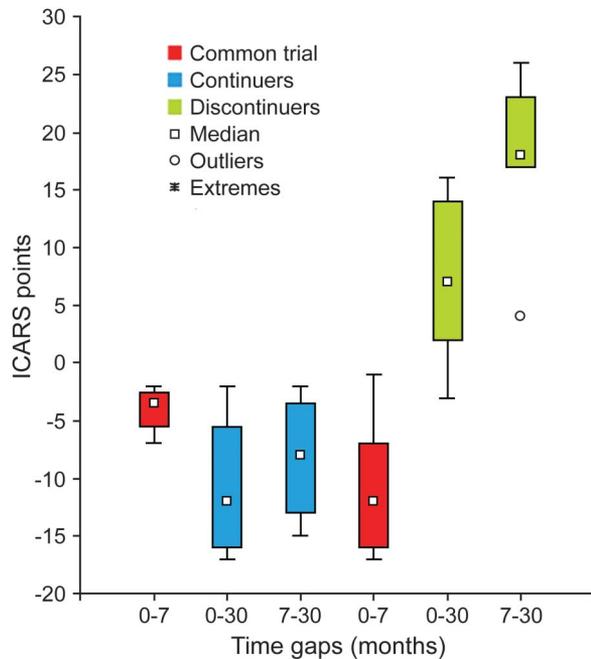
The trend of International Cooperative Ataxia Rating Scale (ICARS) score variations (with SD) in 4 patients with ataxia-telangiectasia who were treated for 30 months with monthly infusions of autologous dexamethasone phosphate–loaded erythrocytes compared with the ICARS scores of 7 patients who discontinued the treatment after the first 6 monthly infusions. 0 (baseline), 7 (after the first 6 infusions), and 30 (after 30 months) represent the scheduled time points for the subsequent assessments. In continuers, 30 is also the total number of infusions before the last evaluation. In red is the common trial (0–7) for all 11 patients, in blue are the ICARS score variations in 4 continuers (0–30; 7–30), and in green are the ICARS score variations in 7 discontinuers (0–30; 7–30). The increase of the score reflects the worsening of ataxia.

RESULTS The table summarizes the clinical response to the treatment at the end of the first 6 months (11 patients) and after 24 further months in continuers (4 male patients) and discontinuers (7 patients; 2 male, 5 female). The mean DSP incorporation during both the original and extended trials is also shown. At the end of the first 6-month trial, ICARS and VABS scores of continuers and discontinuers were ICARS 52.2 ± 7.1 vs 43 ± 2.9 and VABS 6 ± 2.3 vs 8.2 ± 0.6, respectively.

After 30 months, the patients in the extended study experienced an improvement in ICARS score of 10.7 points (SD 6.75; range 2–17) with respect to the score obtained at basal clinical evaluation. Conversely, the ICARS score of the 7 patients who discontinued the treatment worsened by 6.7 points (SD 6.5; range +3–16) (Mann-Whitney $U = 1.00$; $z = 2.45$; $p = 0.013803$). Moreover, all continuers and only 1 of 7 discontinuers improved their ICARS score (Fisher exact test, $p = 0.0152$). Correspondingly, VABS average age improved by 2.10 years (SD 2.4 years; range 9 months–6.1 years) and by 0.9 years (SD 1 year; range −1.6 to +1.10 years) in continuers and discontinuers,

respectively (Mann-Whitney $U = 5.50$; $z = 1.60$; $p = 0.10$). Moreover, ICARS scores at the end of the first 6-month trial and after 30 months improved by 8.2 points (SD 5.8; range 2–17) in the continuers and worsened by 17.8 points (SD 6.9, range −4–26) in the discontinuers (Mann-Whitney $U = 0.00$; $z = 2.64$; $p = 0.008$). Ataxia improved in 4 of 4 continuers and in 1 of 7 discontinuers (Fisher exact test, $p = 0.015$) (figure). Concomitant variations in VABS among continuers and discontinuers were 2.6 years (SD 2.3 years; range 0.5–4.9 years) and −1.7 years (SD 11 months; range −4 months +2.9 years), respectively (Mann-Whitney $U = 0.00$; $z = 2.64$; $p = 0.008$).

No steroid-dependent adverse reactions were observed during the entire treatment period. Standard laboratory tests, physical examination, vital signs, and ECG were all normal. Additional laboratory parameters such as cholesterol (total, high-density lipoprotein, and low-density lipoprotein), glycosylated hemoglobin (HbA1c), and blood and urinary cortisol were all within the normal range. CD4$^+$ lymphocyte count remained stable during the treatment period but below the normal range for 3 of the 4 patients. Blood α-fetoprotein levels increased in all patients, regardless of the treatment they underwent.

DISCUSSION Long-term corticosteroid administration is effective in slowing the progression of Duchenne muscular dystrophy.[10] We recently reported a statistically significant improvement in ataxic symptoms with DSP-loaded red cells in a cohort of young patients with AT.[6] Although this new treatment did not have the usual side effects of chronic steroid treatment, it was apparently less effective then oral betamethasone treatment, which resulted in a median improvement of 13 points on the ICARS after a month of therapy.[5] Present data are more encouraging, showing that after 30 months of treatment the patients experienced an improvement of 10.7 points in mean ICARS score and 3 years in VABS average age with respect to their basal condition. Conversely, the ICARS score of the discontinuers at 30 months had worsened by 6.7 points and the VABS average age had improved just 0.9 years. The gap of 26 ICARS points between continuers and discontinuers at 30 months with respect to the score obtained by the 2 groups after the first 6 months of treatment is an important finding.

Albeit preliminary, the present results are important: 2.5 years is a time span long enough to disclose the progressive neurologic impairment that characterizes the natural clinical history of AT.[1,2] The fact that the discontinuers experienced a decline of their neurologic conditions once steroids were interrupted would suggest a symptomatic effect of the treatment rather than a plastic change involving the basic pathogenetic

mechanism of the disease. Otherwise, the persistent clinical improvement we observed so far in the continuers conflicts with this hypothesis.

Several methodologic limitations (lack of blinding and placebo in the treatment assignation, small size of the sample, lack of external clinical criteria for patient enrollment) affect our study, which was based on an empiric post hoc observation. The lack of AT-dedicated rating scales is a further intrinsic methodologic limitation. Nevertheless, in the face of such a dramatic, untreatable disease, any clinical observation suggesting a potential treatment deserves to be considered.

Whatever the biological effect of steroids in AT, present data support the view that a protracted treatment may delay and perhaps bias the natural course of the disease. Moreover, the delivery system we adopted was confirmed to be safe and well-tolerated, since after this extended period of time no side effects could be detected. A large-scale controlled study on protracted steroid treatment by DSP loaded into erythrocytes in patients with AT is mandatory in order to verify these preliminary data.

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